UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, DC 20460

OFFICE OF
CHEMICAL SAFETY
AND POLLUTION
PREVENTION

June 24, 2014

MEMORANDUM

SUBJECT: Efficacy Review for B-CAP 35 Antimicrobial Agent;
EPA Reg. No. 72372-1;
DP Barcode: D415959

FROM: Karen M. Hill, Ph.D.
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TO: Marshall Swindell RM33/ Zebora Johnson
Regulatory Management Branch I
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APPLICANT: FMC Corporation, Peroxygens Division
1735 Market Street
Philadelphia, PA 19103

Formulation from the Label:

Active Ingredient(s):	% by wt.
Hydrogen Peroxide.....	35%
Inert Ingredients.....	65%
Total.....	100%

I. BACKGROUND:

The product, B-Cap 35 Antimicrobial Agent (EPA Reg. No. 72312-1), is an EPA-approved sterilant for use in treating enclosures up to 37 cubic feet. The applicant requested to amend the registration of the product to add new equipment and additional directions for use claims. The previously approved equipment was Bioquell Hydrogen Peroxide Vapor generator for use with containers of 500 mL or greater. The registrant is seeking to add claims for use with Bioquell QUBE specifically designed for enclosure less than 37 cubic feet with use of 150 mL containers. The study was conducted at Wickmam Laboratories, located at Hoeford Point Barwell Lane Gosprort Hampshire PO13 0AU UK.

This data package contained a letter from the applicant's representative to EPA dated February 25, 2014, EPA Form 8570-4 (CSF), one study (MRID 493246-01), and a Statement of No Data Confidentiality Claims.

II. USE DIRECTIONS:

Directions on the proposed label provide the following information regarding use of the product as a rapid gassing HPV generator using Bioquell QUBE:

The hydrogen peroxide vapor is intended for use as a sterilant in treating enclosures up to 37 cubic feet. Use this product for sterilization as instructed in the Bioquell Use Manual. This product may not be used on food-contact surface unless followed by potable water rinse.

For Enclosures Up to 16 Cubic Feet:

1. Ensure that all porous and non-porous surfaces are visibly clean and free from gross organic contamination.
2. Add B-Cap 35 Antimicrobial Agent according to the Use Manual Instructions.
3. Apply B-Cap 35 Antimicrobial Agent at an injection rate of 6g/minute until 26g has been injected.
4. Allow vapor to remain for a minimum of 10 minutes.
5. Aerate the chamber using hydrogen peroxide vapor is at or below 1.0 ppm. See the Bioquell Use Manual for complete instructions for aeration and recommended methods of measuring hydrogen peroxide vapor.

For Enclosure Greater than 16 Cubic Feet to a Maximum of 37 Cubic Feet:

1. Ensure that all porous and non-porous surfaces are visibly clean and free from gross organic contamination.
2. Add B-Cap 35 Antimicrobial Agent according to the Use Manual Instructions.
3. Apply B-Cap 35 Antimicrobial Agent at an injection rate of 4g/minute until 120g has been injected.
4. Allow vapor to remain for a minimum of 70 minutes.
5. Aerate the chamber using hydrogen peroxide vapor is at or below 1.0 ppm. See the Bioquell Use Manual for complete instructions for aeration and recommended methods of measuring hydrogen peroxide vapor.

III. AGENCY STANDARDS FOR PROPOSED CLAIMS:

Sterilants for Porous and Non-Porous Surfaces within Sealed Enclosures and Vehicles (for Bacterial Spores Known to be Highly Resistant to Sterilants and Disinfectants):

The effectiveness of a sterilant within a sealed enclosure or vehicle may be supported by efficacy data from in-use testing (i.e., field testing) conducted according to an EPA-approved protocol such as "End-Use Protocol for Sterilization of Porous and Non-Porous Surfaces within Sealed Enclosures Using STERIS Vaporized Hydrogen Peroxide (VHP®) Technology" or "End-Use Protocol for Sterilization of Porous and Non-Porous Surfaces within Emergency Vehicle Using STERIS Vaporized Hydrogen Peroxide (VHP®) Technology." Biological Indicators (BIs) must show no growth for the marker organism after 7 days at 55°C. Chemical indicators must show a qualitative color change indicative of hydrogen peroxide exposure. The hydrogen peroxide concentration in the spaces adjacent to the enclosure exterior or vehicle exterior must remain below 1 ppm during the sterilization cycle. Parameters for product application (i.e., temperature, relative humidity, vaporized hydrogen peroxide concentration, contact time) must be met for all four phases of the sterilization cycle.

This large-scale study protocol will be performed using EPA approved sterilization practices for fogging application of the sterilant product. The fogging generation system used in the sterilization process will achieve the airborne test material concentration for the time period required for sterilization. The distribution of the fog will be assisted with fans. The system should be a completely self-contained bi-decontamination system with the ability to dehumidify, generate fog and aerate/decontaminate sealed enclosures. Biological and chemical indicators will be equally distributed throughout the sealed enclosures to allow verification of treatment efficacy. Biological indicators will consist of $\geq 10^6$ *Geobacillus stearothermophilus* spores housed on coupons contained within Tyvek pouches. After treatment, the aeration or decontamination of the sealed enclosures will be performed until the test material is at an acceptable level. Safety monitoring for active ingredient diffusion into adjacent areas will be conducted during the test and in the sealed enclosure after completion of the sterilization process.

IV. COMMENTS ON THE SUBMITTED EFFICACY STUDY:

Certificates of Analysis were submitted for the tested lots. Testing was performed at Bioquell UK Ltd. 52 Royce Close West Portway Andover Hampshire SP10 3TS and at FMC Bayport Texas. The concentration of the active ingredient Hydrogen Peroxide in each tested lot is given below.

Lot	Bioquell H_2O_2 Concentration	FMC H_2O_2 Concentration
A312082913	34%	34.2%
A312082713	34%	34.5%
A312072513	34%	34.5%

Each Lot tested was slightly less than the nominal concentration of 35%.

1. MRID 493246-01 "Sporicidal Activity of B-Cap 35 Antimicrobial Agent As Applied to Bioquell QUBE Rapid Processing Enclosures in Accordance with EPA Requirements," Test Organism: *Geobacillus stearothermophilus* (ATCC 15442), for product B-Cap 35 Antimicrobial Agent, by Susan Wood. Study completion date – November 26, 2013. Study Identification Number 1519.

The study was conducted against *Geobacillus stearothermophilus* (ATCC 15442). Three lots, A312082913, A312082713, and A312072513, of the product were tested using the EPA protocol reference number MRID 490030-01. The product was received ready-to-use. The challenge microorganism was prepared on the biological indicators (BI) at 1.23 X 10^6 mean CFU per carrier disc. The microorganism challenged BI's Lot number is 13171 and Part Number is TD078-1000. The carrier disc was a stainless steel carrier. There was not any evidence of contaminants using standard plating techniques. The culture was incubated at 57.5 ± 2.5°C for 7 days. The D-value for the test organism was 1.25 minutes defined as D-value cycle as per method BI-003. The test material included Bioquell QUBE Rapid Processing Enclosure 0.5 m^3 (16ft^3), Serial number 200000QB0000, System number 000000000PV HPV Chamber only and Bioquell QUBE Rapid Processing Enclosure 1.1 m^3 (37ft^3), Serial number 200000QB0000, System number 000000000PV HPV Complete System Cycle. The enclosures were pre-cleaned removing of visible dirt and waste substances and manufactured from hard non-porous materials. The enclosures contained the following items to represent hard and soft non-porous surfaces found in use conditions: 50 mL snap case glass vials (100 mm X 30 mm), 20 mm silicone stoppers, 12 mL poly propylene sample vials with caps, PTFE stirring rods (8 mm X 150 mm), and sterile graduated pipettes (packaged). The number of biological indicators per chamber is given below.

Lab Reference	Chamber	Cycle	No. of BI's
0013046/1	Bioquell QUBE Rapid Processing Enclosure 0.5 m^3	1	27
0013046/2	Bioquell QUBE Rapid Processing Enclosure 0.5 m^3	2	27
0013046/3	Bioquell QUBE Rapid Processing Enclosure 0.5 m^3	3	27
0013046/4	Bioquell QUBE Rapid Processing Enclosure 1.1 m^3	1	39
0013046/5	Bioquell QUBE Rapid Processing Enclosure 1.1 m^3	2	39
0013046/6	Bioquell QUBE Rapid Processing Enclosure 1.1 m^3	3	39

The equipment was located in a room with temperature at 22 ± 2°C and 50 ± 5% relative humidity. The BI's were placed throughout the Bioquell QUBE enclosure and did not touch the surface of the material it was being hooked to. A small hole in their Tyvek pouch allowed them to be hung on hanging hooks located throughout the enclosure. Three (3) additional BI's were placed within a sealed container prior to each test run for the post-treatment carrier quantification. The cycle parameters are given below.

Cycle Parameters Bioquell QUBE up to 0.5 m^3 (16ft^3):
Gassing Phase = 6.0g/min. injection until 26.0g had been injected
Dwell phase = 0.0g for 10 minutes
Aeration = to 1 ppm or less

Cycle Parameters Bioquell QUBE up to 1.1 m^3 ($37ft^3$):
Gassing Phase = 4.0g/min. injection until 120g had been injected
Dwell phase = 0.0g for 70 minutes
Aeration = to 1 ppm or less

At the end of the defined decontamination cycle, prior to opening the Bioquell QUBE enclosure, the Tyvek pouches containing the BI's were collected from the locations one at a time. Each exposed carrier was removed and transferred to a labelled 10 mL volume of Tryptic Soy Broth. For pre-treatment and post-treatment controls (performed within 24 hours of return), each carrier was transferred to a separate 10 mL volume of Peptone and 0.1% Tween (PET 0.1%) and incubated for 15 minutes prior to being sonicated for 15 minutes. Dilutions were prepared from each sonicated tube. One (1) mL was added to 99 ml of PET 0.1% for dilution 1. After mixing 1 mL of dilution 1, it was added to 9 mL of PET 0.1% for dilution 2. Dilution 2 was heat shocked for 15 minutes at 95-100°C. Afterwards, the tubes were cooled in an ice bath and vortexed. Triplicate 0.5 mL and 1 mL aliquots were plated on Tryptic Soy Agar and incubated for 48 hours at 55-60°C and the number of colonies were counted on each plate and the number of spores per BI calculated. The exposed tested carrier broths and associated control broths were incubated for up to 8 days at 55-60°C. The broths were observed every 1-2 days for signs of growth (turbidity). Controls included pre-treatment and post-treatment carrier quantification, viability, and exposed and unexposed media positive and negative controls.

V. RESULTS:

Organism: *Geobacillus stearothermophilus* Contact (Dwell) Time: **10 minutes or # 70 minutes					
Lab Reference	**Chamber**	**Lot**	**No. of BI's demonstrating growth/ Total No. of BI's tested**	**Pre-Cycle Enumeration Control cfu/disc**	**Post Cycle Enumeration Control cfu/disc**
0013046/1	Bioquell QUBE Rapid Processing Enclosure 0.5 m^3 **	A312082913	0/27	Day 1 2.1 X 10^6	2.0 X 10^6
0013046/2	Bioquell QUBE Rapid Processing Enclosure 0.5 m^3 **	A312082713	0/27	Day 1 2.1 X 10^6	1.7 X 10^6
0013046/3	Bioquell QUBE Rapid Processing Enclosure 0.5 m^3 **	A312072513	0/27	Day 3 1.7 X 10^6	1.9 X 10^6
0013046/4	Bioquell QUBE Rapid Processing Enclosure 1.1 m^3 #	A312082913	0/39	Day 1 2.1 X 10^6	1.8 X 10^6
0013046/5	Bioquell QUBE Rapid Processing Enclosure 1.1 m^3 #	A312082713	0/39	Day 2 1.5 X 10^6	1.7 X 10^6
0013046/6	Bioquell QUBE Rapid Processing Enclosure 1.1 m^3 #	A312072513	0/39	Day 2 1.5 X 10^6	1.5 X 10^6

VI. CONCLUSIONS:

1. The submitted efficacy data does support the use of the product, B-Cap 35 Antimicrobial Agent, as a sterilant on hard, non-porous surfaces with use of the Bioquell QUBE rapid gassing HPV equipment for the indicated conditions:

Cycle Parameters Bioquell QUBE up to 0.5 m^3 ($16ft^3$):
Gassing Phase = 6.0g/min. injection until 26.0g had been injected
Dwell phase = 0.0g for 10 minutes
Aeration = to 1 ppm or less

Cycle Parameters Bioquell QUBE up to 1.1 m^3 ($37ft^3$):
Gassing Phase = 4.0g/min. injection until 120g had been injected
Dwell phase = 0.0g for 70 minutes
Aeration = to 1 ppm or less

Killing was observed on the required number of biological indicators tested against the required number of product lots. The biological indicator population was $\geq 1 \times 10^6$. Each pre-treatment carrier quantification was $\geq 1 \times 10^6$ and each post-treatment carrier quantification was within 1 log of the average result of the pre-treatment carrier quantification. Viability controls were positive for growth. The positive exposed media control showed growth and the negative exposed and unexposed media controls did not show growth.

VII. RECOMMENDATIONS:

1. The below label claims for the product B-Cap 35 Antimicrobial Agent using Bioquell QUBE rapid gassing HPV equipment are **acceptable**:

Cycle Parameters Bioquell QUBE up to 0.5 m^3 ($16ft^3$):
Gassing Phase = 6.0g/min. injection until 26.0g had been injected
Dwell phase = 0.0g for 10 minutes
Aeration = to 1 ppm or less

Cycle Parameters Bioquell QUBE up to 1.1 m^3 ($37ft^3$):
Gassing Phase = 4.0g/min. injection until 120g had been injected
Dwell phase = 0.0g for 70 minutes
Aeration = to 1 ppm or less